

Application No.: 10/717,677  
Response dated: February 28, 2007  
Reply to Office Action dated: November 29, 2006

**Amendments to the Claims:**

This listing of claims will replace all prior versions, and listings, of claims in the application.

**Listing of Claims:**

1. (withdrawn- currently amended) A method of culturing mammalian embryonic stem (ES) cells with reduced differentiation comprising:
  - a) growing the cells in culture on a flexible solid porous matrix without conditioned media and in the absence of fibroblast feeder cells; and
  - b) applying an effective amount of periodic strain ~~to stretch on~~ the flexible matrix to stretch the matrix and the cells thereon, such that the cells proliferate and exhibit reduced differentiation relative to cells not subjected to periodic strain.
2. canceled.
3. (withdrawn) The method of Claim 1 wherein the cells are human embryonic stem cells.
4. (withdrawn) The method of Claim 1 wherein the cell differentiation is eliminated.
5. (withdrawn) The method of Claim 1 wherein the cells are grown on Matrigel™ using BioFlex® untreated culture plates.
6. (withdrawn) The method of Claim 1 wherein the cells are grown without the presence of cross-species biological material.
7. (withdrawn) The method of Claim 1 wherein the flexible matrix is Matrigel™.

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8. (withdrawn) The method of Claim 1 wherein the strain is mechanically produced.

9. (withdrawn) The method of Claim 1 wherein the flexible matrix is stretched using vacuum pressure.

10. (withdrawn) The method of Claim 1 wherein the strain exerted on the flexible matrix is at least about 5%.

11. (withdrawn) The method of Claim 1 wherein the flexible matrix undergoes at least about 6 stretches per minute.

12. (withdrawn) The method of Claim 1 wherein the mechanical strain is from oscillatory stretching of the flexible matrix surface.

13. (currently amended) A cell culture composition comprising:  
mammalian embryonic stem (ES) cells in culture without conditioned media or fibroblast feeder cells; grown on  
a flexible solid porous matrix, wherein the cells are on the matrix; and  
an apparatus for applying without conditioned media or fibroblast feeder cells,  
~~wherein an effective amount of periodic strain is applied to stretch on the flexible matrix to stretch the matrix and the cells thereon, such that the cells proliferate and exhibit reduced differentiation relative to cells not subjected to periodic strain.~~

14. canceled.

15. (previously presented) The culture of Claim 13 wherein the cells are human embryonic stem cells.

16. (previously presented) The culture of Claim 13 wherein the cell differentiation is eliminated.

17. (previously presented) The culture of Claim 13 wherein the cells are grown on Matrigel™ using BioFlex® untreated culture plates.

18. (previously presented) The culture of Claim 13 wherein the cells are grown without the presence of cross-species biological material.

19. (previously presented) The culture of Claim 13 wherein the flexible solid porous matrix is Matrigel™.

20. (previously presented) The culture of Claim 13 wherein the strain is mechanically produced.

21. (previously presented) The culture of Claim 13 wherein the flexible matrix is stretched using vacuum pressure.

22. (previously presented) The culture of Claim 13 wherein the mechanical strain is from oscillatory stretching of the flexible matrix surface.

23. (previously presented) The culture of Claim 13 wherein the strain exerted on the flexible matrix is at least about 5%.

24. (previously presented) The culture of Claim 13 wherein the flexible matrix undergoes at least about 6 stretches per minute.

25. (new) A method of culturing mammalian embryonic stem (ES) cells with reduced differentiation comprising:

- a) growing the cells in culture on a flexible solid porous matrix without conditioned media and in the absence of fibroblast feeder cells; and
- b) applying an effective amount of periodic strain on the cells, such that the cells proliferate and exhibit reduced differentiation relative to cells not subjected to periodic strain.